Week 8 of Introduction to Biological System Design

Feedforward Loops

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Pre-requisite: To get the best out of this notebook, make sure that you have basic understanding of ordinary differential equations (ODE) and Hill functions to model gene regulatory effects. For more information on ODE modeling you may refer to any standard book on engineering math and BFS for more information on Hill functions. You can learn more about how to numerically simulate ODEs deterministically from the week3_intro_ode.ipynb notebook. Further, it is also assumed that you have a working knowledge of gene expression processes, use of Hill functions for gene regulation, and biological system motifs. Computational examples with Hill functions are discussed in week4_hill_functions.ipynb whereas design choices underlying biological motifs are shown in week6_system_analysis.ipynb. This notebook builds on the code discussed in week6_system_analysis.ipynb to analyze feedforward loop motifs.

Disclaimer: Concepts demonstrated in this notebook have been inspired from the discussion on feedforward loops in Alon and Biocircuits Lecture by Elowitz and Bois.

In [236…

```
# To plot heatmaps in this notebook, you may need to
# install a package called "seaborn"
# To install seaborn, run the following command
# (or install the package "seaborn" using the Anaconda Navigator search)

# !pip install seaborn
```

Coherent Feedforward Loops (CFFL)

Consider the motif where X --> Y --> Z and X --> Z indirectly as well.

C1-FFL with AND logic

In [209…

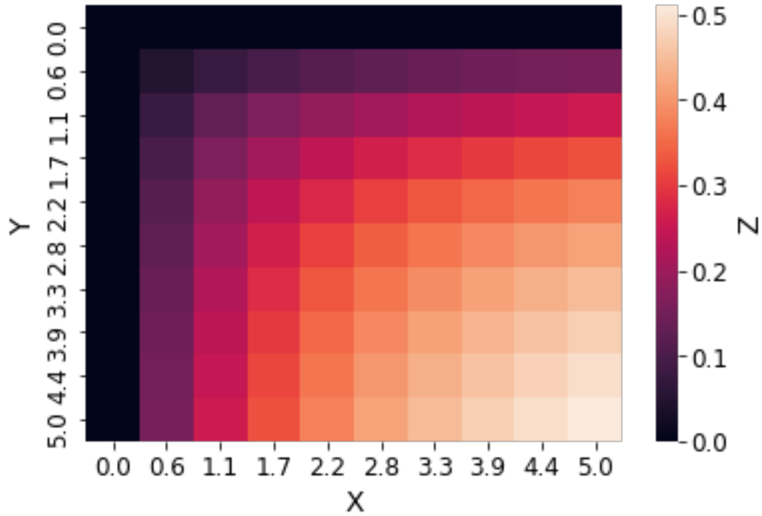
```
def c1_ffl_and(x,t,*args):
    """
    ODE model for C1-FFL with AND logic.
    """
    k, n_X, K_X, n_Y, K_Y, d_Z = args
    X, Y, Z = x
    dZ_dt = k * (X**n_X)/(K_X**n_X + X**n_X) *\
            (Y**n_Y)/(K_Y**n_Y + Y**n_Y) - d_Z * Z
    # Since X and Y don't change, the rate of change
    # of X and Y is equal to zero. We are only modeling
    # rate of change of Z.
    return np.array([0, 0, dZ_dt])
```

In [210…

```
import numpy as np
from scipy.integrate import odeint

X = np.linspace(0, 5, 10)
Y = np.linspace(0, 5, 10)
timepoints = np.linspace(0,100,10)
Z_ss = np.zeros((len(X),len(Y)))

# parameters:
k = 1
n_X = 1
K_X = 2
n_Y = 1
K_Y = 2
d_Z = 1

for i, x0 in enumerate(X):
    for j, y0 in enumerate(Y):
        initial_condition = np.array([x0,y0,0])
        solution = odeint(c1_ffl_and, y0 = initial_condition,
                          t = timepoints,
                          args = (k, n_X, K_X, n_Y, K_Y, d_Z))
        # Store steady-state value
        Z_ss[i,j] = solution[:,2][-1]
```

In [211…

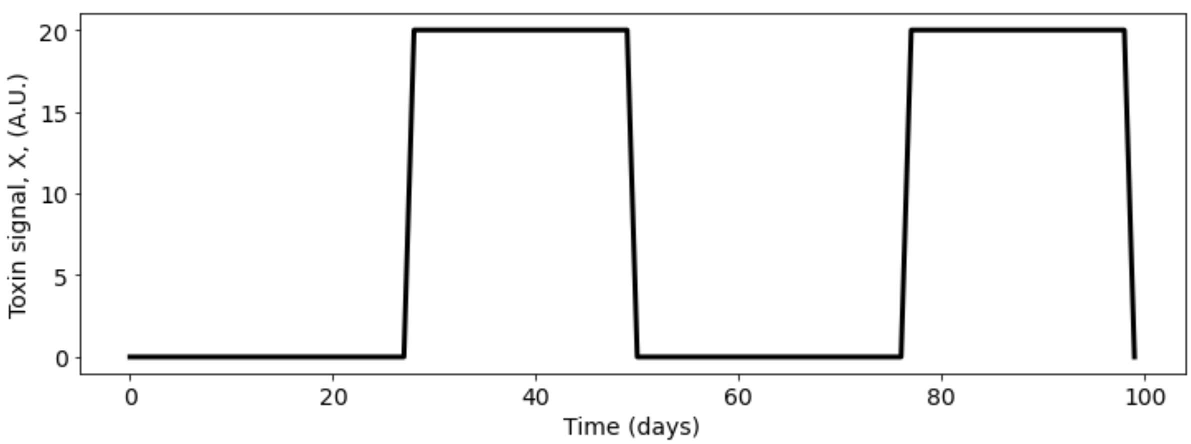
```
import seaborn as sn
ax = sn.heatmap(Z_ss, xticklabels = np.around(X,1),
                yticklabels = np.around(Y,1))
ax.tick_params(labelsize = 12)
cbar_ax = ax.figure.axes[-1]
cbar_ax.tick_params(labelsize = 12)
cbar_ax.set_ylabel('Z', fontsize = 14)
ax.set_xlabel('X', fontsize = 14)
ax.set_ylabel('Y', fontsize = 14);
```



C1-FFL with AND logic exhibits delayed response

In [212…

```
from scipy import signal
timepoints = np.linspace(0, 100, 100, endpoint = True)
max_toxin_value = 20 #arbitrary units
toxin_signal = max_toxin_value*np.ones_like(timepoints) *\
-1*signal.square(2*np.pi*2*timepoints, duty = 0.55)
for i, s in enumerate(toxin_signal):
    if s < 0:
        toxin_signal[i] = 0
fig, ax = plt.subplots(figsize = (12,4))
ax.plot(toxin_signal, color = 'black', lw = 3)
ax.set_xlabel('Time (days)', fontsize = 14)
ax.set_ylabel('Toxin signal, X, (A.U.)', fontsize = 14)
ax.tick_params(labelsize = 14)
```



In [213…

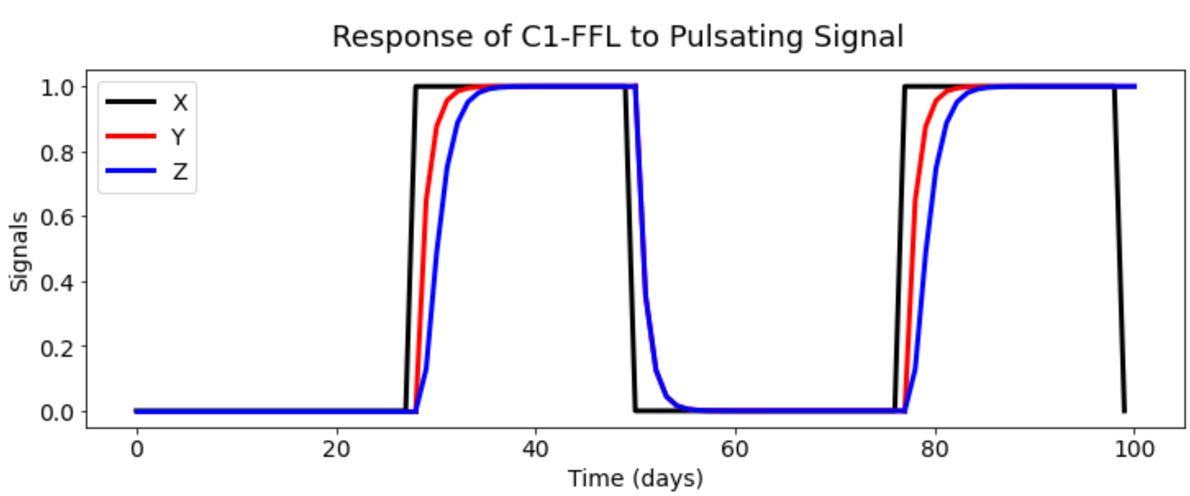
```
def c1_ffl_and(x,t,*args):
    """
    ODE model for C1-FFL with AND logic.
    """
    k_Y, k_Z, n_X, K_X, n_Y, K_Y, d_Y, d_Z = args
    X, Y, Z = x
    dY_dt = k_Y * (X**n_X)/(K_X**n_X + X**n_X) - d_Y * Y
    dZ_dt = k_Z * (X**n_X)/(K_X**n_X + X**n_X) *\
            (Y**n_Y)/(K_Y**n_Y + Y**n_Y) - d_Z * Z
    # Since X is fixed input, it doesn't change.
    # the rate of change
    # of X is equal to zero. We are only modeling
    # rate of change of Y and Z.
    return np.array([0, dY_dt, dZ_dt])
```

In [214…

```python
fig, ax = plt.subplots(figsize = (12,4))
fig.suptitle('Response of C1-FFL (AND logic) to Pulsating Signal',
             fontsize = 18);
# parameters:
k_Y = 1
k_Z = 1
n_X = 3
K_X = 1
n_Y = 3
K_Y = 1
d_Y = 1
d_Z = 1

# Normalize the values
def normalize(solution):
    """
    Normalize by maximum value in the odeint solution
    except when the values are zero, to avoid division by zero.
    """
    normalized_solution = np.zeros_like(solution.T)
    for i, val_array in enumerate(solution.T):
        max_value = np.max(val_array)
        for j, val in enumerate(val_array):
            if max_value == 0:
                normalized_solution[i, j] = val
            else:
                normalized_solution[i, j] = val/max_value
    return normalized_solution.T

# Plot X
ax.plot(toxin_signal/np.max(toxin_signal), color = 'black',
        lw = 3, label = 'X')

# For X = 0
previous_time = 0
array_nonzero = np.where(toxin_signal != 0)[0]
next_time = array_nonzero[0]
t_solve = np.linspace(previous_time, next_time,
                      next_time - previous_time, endpoint = True)
solution = odeint(c1_ffl_and, y0 = np.array([0, 0, 0]),
                  t = t_solve,
                  args = (k_Y, k_Z, n_X, K_X, n_Y,
                          K_Y, d_Y, d_Z ))
normalized_solution = normalize(solution)
ax.plot(t_solve, normalized_solution[:,1], 'r', lw = 3, label = 'Y')
ax.plot(t_solve, normalized_solution[:,2], 'b', lw = 3, label = 'Z')

# For X = max_toxin_value
previous_time = next_time
array_zero = np.where(toxin_signal == 0)[0]
next_time = array_zero[np.where(array_zero > previous_time)][0]
t_solve = np.linspace(previous_time,next_time,
                      next_time - previous_time, endpoint = True)
solution = odeint(c1_ffl_and, y0 = np.array([max_toxin_value, 0, 0]),
                  t = t_solve,
                  args = (k_Y, k_Z, n_X, K_X, n_Y,
```

```python
                                K_Y, d_Y, d_Z ))
normalized_solution = normalize(solution)
ax.plot(t_solve, normalized_solution[:,1], 'r', lw = 3)
ax.plot(t_solve, normalized_solution[:,2], 'b', lw = 3)

y_ss = normalized_solution[:,1][-1]
z_ss = normalized_solution[:,2][-1]
# For X = 0 again
previous_time = next_time
array_zero = np.where(toxin_signal != 0)[0]
next_time = array_zero[np.where(array_zero > previous_time)][0]
t_solve = np.linspace(previous_time, next_time,
                      next_time - previous_time, endpoint = True)
solution = odeint(c1_ffl_and, y0 = np.array([0, y_ss, z_ss]),
                  t = t_solve,
                  args = (k_Y, k_Z, n_X, K_X, n_Y,
                          K_Y, d_Y, d_Z ))
normalized_solution = normalize(solution)
ax.plot(t_solve, normalized_solution[:,1], 'r', lw = 3)
ax.plot(t_solve, normalized_solution[:,2], 'b', lw = 3)

# For X = max_toxin_value, again
previous_time = next_time
next_time = int(timepoints[-1]) # last point
t_solve = np.linspace(previous_time, next_time,
                      next_time - previous_time, endpoint = True)
solution = odeint(c1_ffl_and, y0 = np.array([max_toxin_value, 0, 0]),
                  t = t_solve,
                  args = (k_Y, k_Z, n_X, K_X, n_Y,
                          K_Y, d_Y, d_Z ))
normalized_solution = normalize(solution)
ax.plot(t_solve, normalized_solution[:,1], 'r', lw = 3)
ax.plot(t_solve, normalized_solution[:,2], 'b', lw = 3)

ax.set_xlabel('Time (days)', fontsize = 14)
ax.set_ylabel('Signals', fontsize = 14)
ax.tick_params(labelsize = 14)
ax.legend(fontsize = 14)
```

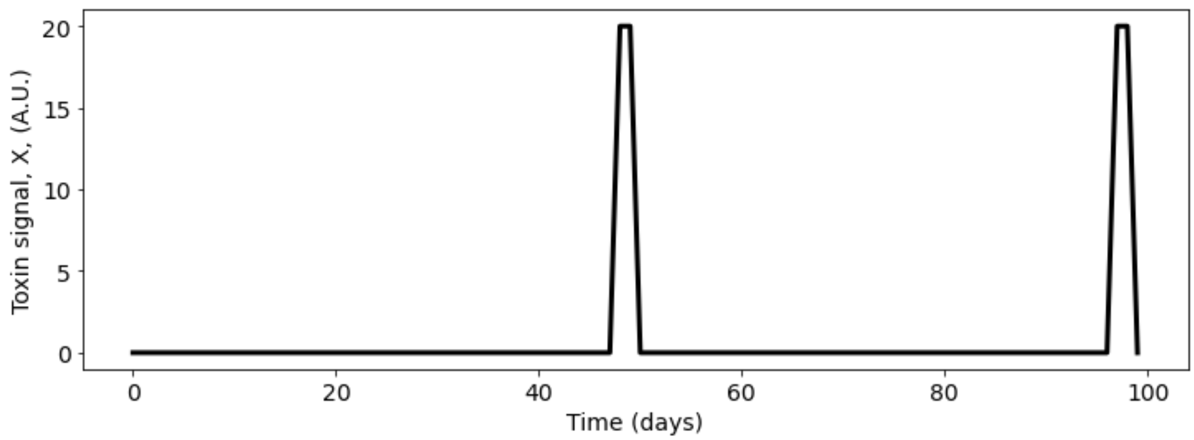
Out[214]:
```
<matplotlib.legend.Legend at 0x1f1f39ecfd0>
```



C1-FFL with AND logic filters short pulses

In [223…

```
from scipy import signal
timepoints = np.linspace(0, 100, 100, endpoint = True)
max_toxin_value = 20 #arbitrary units
toxin_signal = max_toxin_value*np.ones_like(timepoints) *\
-1*signal.square(2*np.pi*2*timepoints, duty = 0.95)
for i, s in enumerate(toxin_signal):
    if s < 0:
        toxin_signal[i] = 0
fig, ax = plt.subplots(figsize = (12,4))
ax.plot(toxin_signal, color = 'black', lw = 3)
ax.set_xlabel('Time (days)', fontsize = 14)
ax.set_ylabel('Toxin signal, X, (A.U.)', fontsize = 14)
ax.tick_params(labelsize = 14)
```

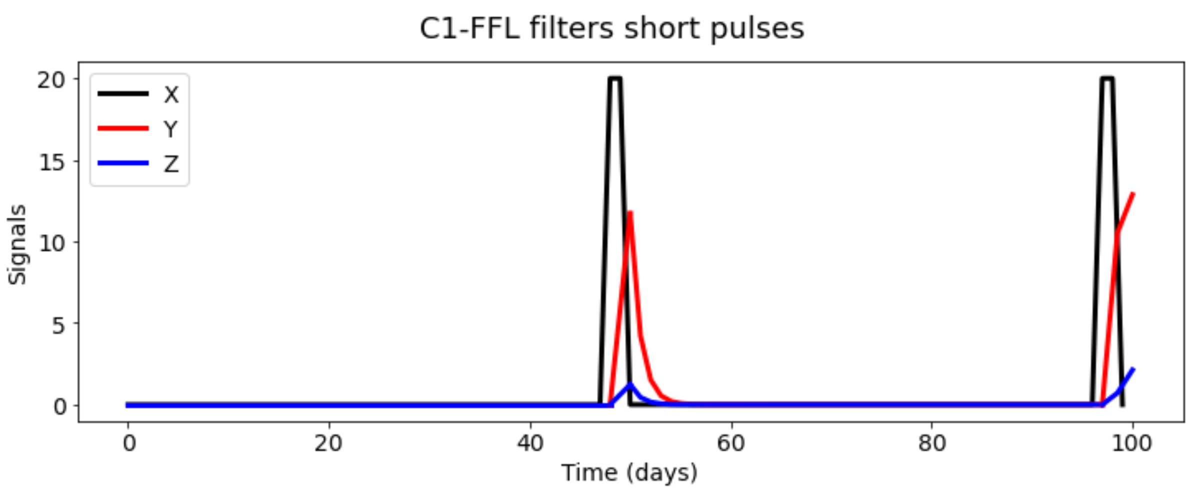


In [225…

```
fig, ax = plt.subplots(figsize = (12,4))
fig.suptitle('C1-FFL filters short pulses',
             fontsize = 18);
# parameters:
k_Y = 40
k_Z = 40
n_X = 3
K_X = 25
n_Y = 3
K_Y = 20
d_Y = 1
d_Z = 1

# Plot X
ax.plot(toxin_signal, color = 'black',
        lw = 3, label = 'X')

# For X = 0
previous_time = 0
array_nonzero = np.where(toxin_signal != 0)[0]
next_time = array_nonzero[0]
t_solve = np.linspace(previous_time, next_time,
                      next_time - previous_time, endpoint = True)
solution = odeint(c1_ffl_and, y0 = np.array([0, 0, 0]),
                  t = t_solve,
                  args = (k_Y, k_Z, n_X, K_X, n_Y,
                          K_Y, d_Y, d_Z ))
ax.plot(t_solve, solution[:,1], 'r', lw = 3, label = 'Y')
ax.plot(t_solve, solution[:,2], 'b', lw = 3, label = 'Z')

# For X = max_toxin_value
previous_time = next_time
array_zero = np.where(toxin_signal == 0)[0]
next_time = array_zero[np.where(array_zero > previous_time)][0]
t_solve = np.linspace(previous_time,next_time,
                      next_time - previous_time, endpoint = True)
solution = odeint(c1_ffl_and, y0 = np.array([max_toxin_value, 0, 0]),
                  t = t_solve,
                  args = (k_Y, k_Z, n_X, K_X, n_Y,
                          K_Y, d_Y, d_Z ))
ax.plot(t_solve, solution[:,1], 'r', lw = 3)
ax.plot(t_solve, solution[:,2], 'b', lw = 3)

y_ss = solution[:,1][-1]
z_ss = solution[:,2][-1]
# For X = 0 again
previous_time = next_time
array_zero = np.where(toxin_signal != 0)[0]
next_time = array_zero[np.where(array_zero > previous_time)][0]
t_solve = np.linspace(previous_time, next_time,
                      next_time - previous_time, endpoint = True)
solution = odeint(c1_ffl_and, y0 = np.array([0, y_ss, z_ss]),
                  t = t_solve,
                  args = (k_Y, k_Z, n_X, K_X, n_Y,
                          K_Y, d_Y, d_Z ))
ax.plot(t_solve, solution[:,1], 'r', lw = 3)
```

```
ax.plot(t_solve, solution[:,2], 'b', lw = 3)

# For X = max_toxin_value, again
previous_time = next_time
next_time = int(timepoints[-1]) # last point
t_solve = np.linspace(previous_time, next_time,
                      next_time - previous_time, endpoint = True)
solution = odeint(c1_ffl_and, y0 = np.array([max_toxin_value, 0, 0]),
                  t = t_solve,
                  args = (k_Y, k_Z, n_X, K_X, n_Y,
                          K_Y, d_Y, d_Z ))
ax.plot(t_solve, solution[:,1], 'r', lw = 3)
ax.plot(t_solve, solution[:,2], 'b', lw = 3)

ax.set_xlabel('Time (days)', fontsize = 14)
ax.set_ylabel('Signals', fontsize = 14)
ax.tick_params(labelsize = 14)
ax.legend(fontsize = 14)
```

Out[225]: <matplotlib.legend.Legend at 0x1f1f4fa6e80>



C1-FFL with OR logic

In [226…

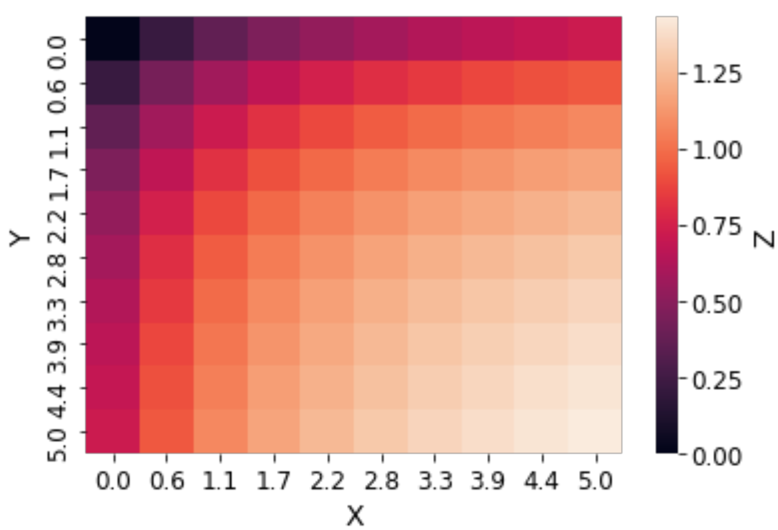
```
def c1_ffl_or(x,t,*args):
    """
    ODE model for C1-FFL with AND logic.
    """
    k, n_X, K_X, n_Y, K_Y, d_Z = args
    X, Y, Z = x
    dZ_dt = k * ((X**n_X)/(K_X**n_X + X**n_X) +\
            (Y**n_Y)/(K_Y**n_Y + Y**n_Y)) - d_Z * Z
    # Since X and Y don't change, the rate of change
    # of X and Y is equal to zero. We are only modeling
    # rate of change of Z.
    return np.array([0, 0, dZ_dt])
```

In [227…

```
X = np.linspace(0, 5, 10)
Y = np.linspace(0, 5, 10)
timepoints = np.linspace(0,100,10)
Z_ss = np.zeros((len(X),len(Y)))

# parameters:
k = 1
n_X = 1
K_X = 2
n_Y = 1
K_Y = 2
d_Z = 1

for i, x0 in enumerate(X):
    for j, y0 in enumerate(Y):
        initial_condition = np.array([x0,y0,0])
        solution = odeint(c1_ffl_or, y0 = initial_condition,
                          t = timepoints,
                          args = (k, n_X, K_X, n_Y, K_Y, d_Z))
        # Store steady-state value
        Z_ss[i,j] = solution[:,2][-1]
```

In [228…

```
import seaborn as sn
ax = sn.heatmap(Z_ss, xticklabels = np.around(X,1),
                yticklabels = np.around(Y,1))
ax.tick_params(labelsize = 12)
cbar_ax = ax.figure.axes[-1]
cbar_ax.tick_params(labelsize = 12)
cbar_ax.set_ylabel('Z', fontsize = 14)
ax.set_xlabel('X', fontsize = 14)
ax.set_ylabel('Y', fontsize = 14);
```



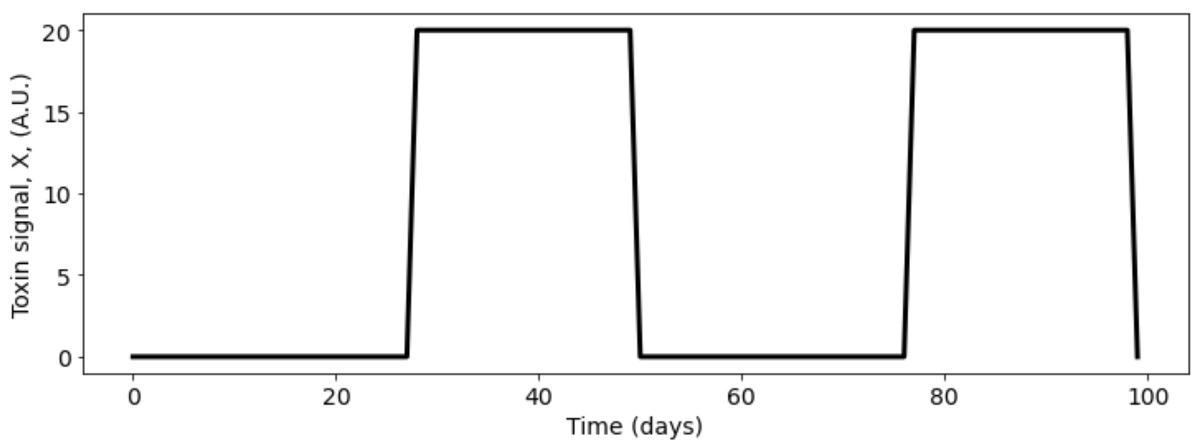
C1-FFL with OR logic exhibits delayed response

In [231…

```
def c1_ffl_or(x,t,*args):
    """
    ODE model for C1-FFL with OR logic.
    """
    k_Y, k_Z, n_X, K_X, n_Y, K_Y, d_Y, d_Z = args
    X, Y, Z = x
    dY_dt = k_Y * (X**n_X)/(K_X**n_X + X**n_X) - d_Y * Y
    dZ_dt = k_Z * (X**n_X)/(K_X**n_X + X**n_X) +\
            (Y**n_Y)/(K_Y**n_Y + Y**n_Y) - d_Z * Z
    # Since X is fixed input, it doesn't change.
    # the rate of change
    # of X is equal to zero. We are only modeling
    # rate of change of Y and Z.
    return np.array([0, dY_dt, dZ_dt])
```

In [232…

```
from scipy import signal
timepoints = np.linspace(0, 100, 100, endpoint = True)
max_toxin_value = 20 #arbitrary units
toxin_signal = max_toxin_value*np.ones_like(timepoints) *\
-1*signal.square(2*np.pi*2*timepoints, duty = 0.55)
for i, s in enumerate(toxin_signal):
    if s < 0:
        toxin_signal[i] = 0
fig, ax = plt.subplots(figsize = (12,4))
ax.plot(toxin_signal, color = 'black', lw = 3)
ax.set_xlabel('Time (days)', fontsize = 14)
ax.set_ylabel('Toxin signal, X, (A.U.)', fontsize = 14)
ax.tick_params(labelsize = 14)
```

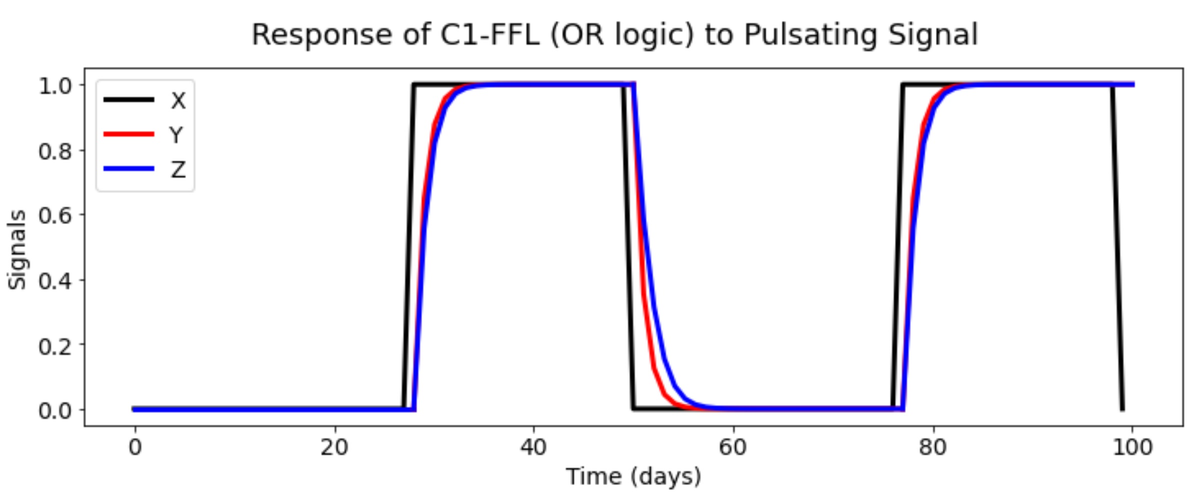


In [235…

```
fig, ax = plt.subplots(figsize = (12,4))
fig.suptitle('Response of C1-FFL (OR logic) to Pulsating Signal',
             fontsize = 18);
# parameters:
k_Y = 1
k_Z = 1
n_X = 1
K_X = 1
n_Y = 1
K_Y = 1
d_Y = 1
d_Z = 1

# Normalize the values
def normalize(solution):
    """
    Normalize by maximum value in the odeint solution
    except when the values are zero, to avoid division by zero.
    """
    normalized_solution = np.zeros_like(solution.T)
    for i, val_array in enumerate(solution.T):
        max_value = np.max(val_array)
        for j, val in enumerate(val_array):
            if max_value == 0:
                normalized_solution[i, j] = val
            else:
                normalized_solution[i, j] = val/max_value
    return normalized_solution.T

# Plot X
ax.plot(toxin_signal/np.max(toxin_signal), color = 'black',
        lw = 3, label = 'X')

# For X = 0
previous_time = 0
array_nonzero = np.where(toxin_signal != 0)[0]
next_time = array_nonzero[0]
t_solve = np.linspace(previous_time, next_time,
                      next_time - previous_time, endpoint = True)
solution = odeint(c1_ffl_or, y0 = np.array([0, 0, 0]),
                  t = t_solve,
                  args = (k_Y, k_Z, n_X, K_X, n_Y,
                          K_Y, d_Y, d_Z ))
normalized_solution = normalize(solution)
ax.plot(t_solve, normalized_solution[:,1], 'r', lw = 3, label = 'Y')
ax.plot(t_solve, normalized_solution[:,2], 'b', lw = 3, label = 'Z')

# For X = max_toxin_value
previous_time = next_time
array_zero = np.where(toxin_signal == 0)[0]
next_time = array_zero[np.where(array_zero > previous_time)][0]
t_solve = np.linspace(previous_time,next_time,
                      next_time - previous_time, endpoint = True)
solution = odeint(c1_ffl_or, y0 = np.array([max_toxin_value, 0, 0]),
                  t = t_solve,
                  args = (k_Y, k_Z, n_X, K_X, n_Y,
```

```
                          K_Y, d_Y, d_Z ))
normalized_solution = normalize(solution)
ax.plot(t_solve, normalized_solution[:,1], 'r', lw = 3)
ax.plot(t_solve, normalized_solution[:,2], 'b', lw = 3)

y_ss = normalized_solution[:,1][-1]
z_ss = normalized_solution[:,2][-1]

# For X = 0 again
previous_time = next_time
array_zero = np.where(toxin_signal != 0)[0]
next_time = array_zero[np.where(array_zero > previous_time)][0]
t_solve = np.linspace(previous_time, next_time,
                      next_time - previous_time, endpoint = True)
solution = odeint(c1_ffl_or, y0 = np.array([0, y_ss, z_ss]),
                  t = t_solve,
                  args = (k_Y, k_Z, n_X, K_X, n_Y,
                          K_Y, d_Y, d_Z ))
normalized_solution = normalize(solution)
ax.plot(t_solve, normalized_solution[:,1], 'r', lw = 3)
ax.plot(t_solve, normalized_solution[:,2], 'b', lw = 3)

# For X = max_toxin_value, again
previous_time = next_time
next_time = int(timepoints[-1]) # last point
t_solve = np.linspace(previous_time, next_time,
                      next_time - previous_time, endpoint = True)
solution = odeint(c1_ffl_or, y0 = np.array([max_toxin_value, 0, 0]),
                  t = t_solve,
                  args = (k_Y, k_Z, n_X, K_X, n_Y,
                          K_Y, d_Y, d_Z ))
normalized_solution = normalize(solution)
ax.plot(t_solve, normalized_solution[:,1], 'r', lw = 3)
ax.plot(t_solve, normalized_solution[:,2], 'b', lw = 3)

ax.set_xlabel('Time (days)', fontsize = 14)
ax.set_ylabel('Signals', fontsize = 14)
ax.tick_params(labelsize = 14)
ax.legend(fontsize = 14)
```

Out[235]: <matplotlib.legend.Legend at 0x1f1e1f91828>



Incoherent Feedforward Loops (IFFL)

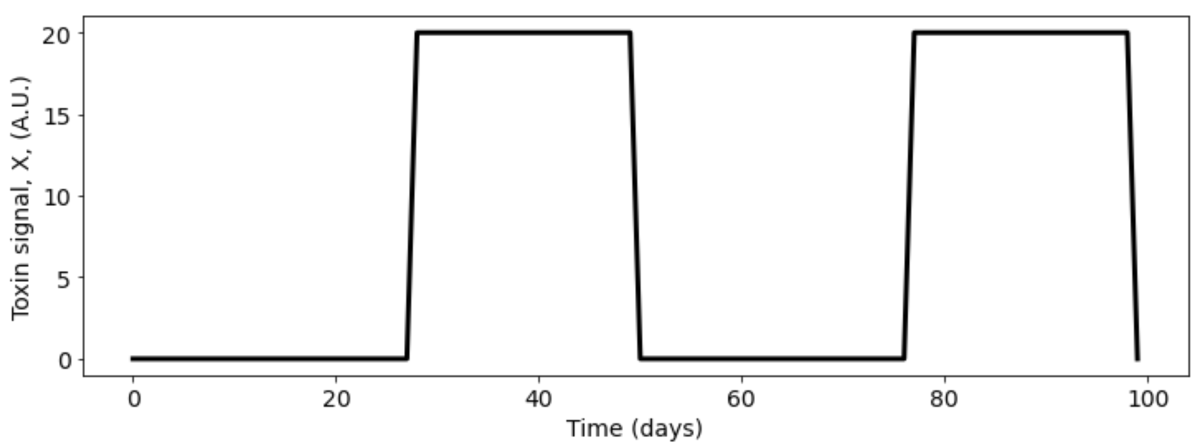
Consider the motif where X --> Y --| Z and X --> Z indirectly as well.

In [240…

```
def i1_ffl(x,t,*args):
    """
    ODE model for I1-FFL.
    """
    k_Y, k_Z, n_X, K_X, n_Y, K_Y, d_Y, d_Z = args
    X, Y, Z = x
    dY_dt = k_Y * (X**n_X)/(K_X**n_X + X**n_X) - d_Y * Y
    dZ_dt = k_Z * (X**n_X)/(K_X**n_X + X**n_X) *\
            (K_Y**n_Y)/(K_Y**n_Y + Y**n_Y) - d_Z * Z
    # Since X is fixed input, it doesn't change.
    # the rate of change
    # of X is equal to zero. We are only modeling
    # rate of change of Y and Z.
    return np.array([0, dY_dt, dZ_dt])
```

In [241…

```
from scipy import signal
timepoints = np.linspace(0, 100, 100, endpoint = True)
max_toxin_value = 20 #arbitrary units
toxin_signal = max_toxin_value*np.ones_like(timepoints) *\
-1*signal.square(2*np.pi*2*timepoints, duty = 0.55)
for i, s in enumerate(toxin_signal):
    if s < 0:
        toxin_signal[i] = 0
fig, ax = plt.subplots(figsize = (12,4))
ax.plot(toxin_signal, color = 'black', lw = 3)
ax.set_xlabel('Time (days)', fontsize = 14)
ax.set_ylabel('Toxin signal, X, (A.U.)', fontsize = 14)
ax.tick_params(labelsize = 14)
```

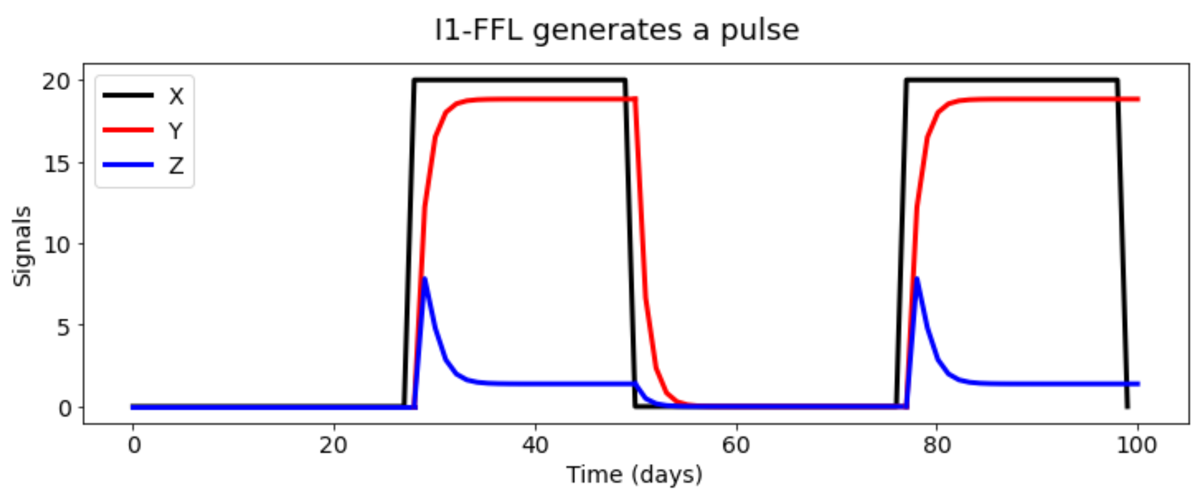


In [252…

```python
fig, ax = plt.subplots(figsize = (12,4))
fig.suptitle('I1-FFL generates a pulse',
             fontsize = 18);
# parameters:
k_Y = 20
k_Z = 20
n_X = 4
K_X = 10
n_Y = 4
K_Y = 10
d_Y = 1
d_Z = 1

# Plot X
ax.plot(toxin_signal, color = 'black',
        lw = 3, label = 'X')

# For X = 0
previous_time = 0
array_nonzero = np.where(toxin_signal != 0)[0]
next_time = array_nonzero[0]
t_solve = np.linspace(previous_time, next_time,
                      next_time - previous_time, endpoint = True)
solution = odeint(i1_ffl, y0 = np.array([0, 0, 0]),
                  t = t_solve,
                  args = (k_Y, k_Z, n_X, K_X, n_Y,
                          K_Y, d_Y, d_Z ))
ax.plot(t_solve, solution[:,1], 'r', lw = 3, label = 'Y')
ax.plot(t_solve, solution[:,2], 'b', lw = 3, label = 'Z')

# For X = max_toxin_value
previous_time = next_time
array_zero = np.where(toxin_signal == 0)[0]
next_time = array_zero[np.where(array_zero > previous_time)][0]
t_solve = np.linspace(previous_time,next_time,
                      next_time - previous_time, endpoint = True)
solution = odeint(i1_ffl, y0 = np.array([max_toxin_value, 0, 0]),
                  t = t_solve,
                  args = (k_Y, k_Z, n_X, K_X, n_Y,
                          K_Y, d_Y, d_Z ))
ax.plot(t_solve, solution[:,1], 'r', lw = 3)
ax.plot(t_solve, solution[:,2], 'b', lw = 3)

y_ss = solution[:,1][-1]
z_ss = solution[:,2][-1]
# For X = 0 again
previous_time = next_time
array_zero = np.where(toxin_signal != 0)[0]
next_time = array_zero[np.where(array_zero > previous_time)][0]
t_solve = np.linspace(previous_time, next_time,
                      next_time - previous_time, endpoint = True)
solution = odeint(i1_ffl, y0 = np.array([0, y_ss, z_ss]),
                  t = t_solve,
                  args = (k_Y, k_Z, n_X, K_X, n_Y,
                          K_Y, d_Y, d_Z ))
ax.plot(t_solve, solution[:,1], 'r', lw = 3)
```

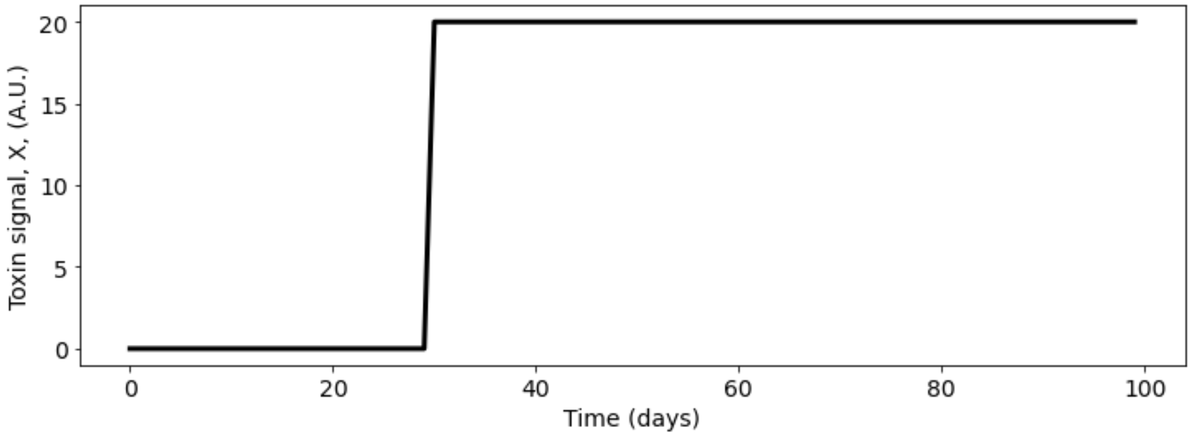
```
ax.plot(t_solve, solution[:,2], 'b', lw = 3)

# For X = max_toxin_value, again
previous_time = next_time
next_time = int(timepoints[-1]) # last point
t_solve = np.linspace(previous_time, next_time,
                      next_time - previous_time, endpoint = True)
solution = odeint(i1_ffl, y0 = np.array([max_toxin_value, 0, 0]),
                  t = t_solve,
                  args = (k_Y, k_Z, n_X, K_X, n_Y,
                          K_Y, d_Y, d_Z ))
ax.plot(t_solve, solution[:,1], 'r', lw = 3)
ax.plot(t_solve, solution[:,2], 'b', lw = 3)

ax.set_xlabel('Time (days)', fontsize = 14)
ax.set_ylabel('Signals', fontsize = 14)
ax.tick_params(labelsize = 14)
ax.legend(fontsize = 14)
```

Out[252]: <matplotlib.legend.Legend at 0x1f1f673d978>



In [284…

```
from scipy import signal
timepoints = np.linspace(0, 100, 100, endpoint = True)
max_toxin_value = 20 #arbitrary units
toxin_signal = max_toxin_value*np.ones_like(timepoints) *\
-1*signal.square(2*np.pi*1*timepoints, duty = 0.3)
for i, s in enumerate(toxin_signal):
    if s < 0:
        toxin_signal[i] = 0
toxin_signal[-1] = 20
fig, ax = plt.subplots(figsize = (12,4))
ax.plot(toxin_signal, color = 'black', lw = 3)
ax.set_xlabel('Time (days)', fontsize = 14)
ax.set_ylabel('Toxin signal, X, (A.U.)', fontsize = 14)
ax.tick_params(labelsize = 14)
```



In [285…

```
def unregulated(x, t, *args):
    k, d = args
    return k - d*x
```

In [292…

```python
fig, ax = plt.subplots(figsize = (12,4))
fig.suptitle('Response of I1-FFL to Pulsating Signal',
             fontsize = 18);
# parameters  (IFFL):
k_Y = 1
k_Z = 1
n_X = 4
K_X = 1
n_Y = 4
K_Y = 1
d_Y = 0.5
d_Z = 0.5

# parameters (unregulated):
k = 1
d = 0.5

# Normalize the values
def normalize(solution):
    """
    Normalize by maximum value in the odeint solution
    except when the values are zero, to avoid division by zero.
    """
    normalized_solution = np.zeros_like(solution.T)
    for i, val_array in enumerate(solution.T):
        max_value = np.max(val_array)
        for j, val in enumerate(val_array):
            if max_value == 0:
                normalized_solution[i, j] = val
            else:
                normalized_solution[i, j] = val/max_value
    return normalized_solution.T

# Plot X
ax.plot(toxin_signal/np.max(toxin_signal), color = 'black',
        lw = 3, label = 'X')

# For X = 0
previous_time = 0
array_nonzero = np.where(toxin_signal != 0)[0]
next_time = array_nonzero[0]
t_solve = np.linspace(previous_time, next_time,
                      next_time - previous_time, endpoint = True)
solution = odeint(i1_ffl, y0 = np.array([0, 0, 0]),
                  t = t_solve,
                  args = (k_Y, k_Z, n_X, K_X, n_Y,
                          K_Y, d_Y, d_Z ))
normalized_solution = normalize(solution)

ax.plot(t_solve, normalized_solution[:,1], 'r', lw = 3, label = 'Y')
ax.plot(t_solve, normalized_solution[:,2], 'b', lw = 3, label = 'Z')

# For X = max_toxin_value
previous_time = next_time
array_zero = np.where(toxin_signal == 0)[0]
next_time = int(timepoints[-1])
```

```
t_solve = np.linspace(previous_time,next_time,
                      next_time - previous_time, endpoint = True)
solution = odeint(i1_ffl, y0 = np.array([max_toxin_value, 0, 0]),
                  t = t_solve,
                  args = (k_Y, k_Z, n_X, K_X, n_Y,
                          K_Y, d_Y, d_Z ))
normalized_solution = normalize(solution)

ax.plot(t_solve, normalized_solution[:,1], 'r', lw = 3)
ax.plot(t_solve, normalized_solution[:,2], 'b', lw = 3)

unreg_solution = odeint(unregulated, y0 = np.array([0]),
                        t = t_solve,
                        args = (k,d))
unreg_normalized_solution = normalize(unreg_solution)
ax.plot(t_solve, unreg_normalized_solution, color = 'orange', lw = 3)

y_ss = normalized_solution[:,1][-1]
z_ss = normalized_solution[:,2][-1]

ax.set_xlabel('Time (days)', fontsize = 14)
ax.set_ylabel('Signals', fontsize = 14)
ax.tick_params(labelsize = 14)
ax.legend(fontsize = 14)
```

Out[292]: <matplotlib.legend.Legend at 0x1f1f516de48>